

# Alternate Host Phenology Affects Survivorship, Growth, and Development of Western Corn Rootworm (Coleoptera: Chrysomelidae) Larvae

PETER GACII CHEGE,<sup>1</sup> THOMAS L. CLARK,<sup>1, 2</sup> AND BRUCE E. HIBBARD<sup>2, 3</sup>

Environ. Entomol. 34(6): 1441–1447 (2005)

**ABSTRACT** The commercial release of transgenic maize with resistance to rootworms incorporated with glyphosate tolerance has become a reality, and questions have arisen regarding the impact of grassy weed phenology on the biology of the western corn rootworm, *Diabrotica virgifera virgifera* LeConte. A study to determine the impact of host plant phenology on the survivorship (percentage larval recovery), growth (dry-weight gain), and development (change in head capsule width) of western corn rootworm larvae was conducted in a split-split-plot randomized complete block design experiment under greenhouse conditions. Six host species were evaluated under eight weekly infestations of 15 neonate western corn rootworm larvae that were sampled for larval recovery after 7, 14, and 21 d, each with five replications. Percentage larval recovery, change in head capsule width and weight gain were significantly impacted by infestation time and host species. Other than from maize, *Zea mays* L., larval survivorship was highest in large crabgrass, *Digitaria sanguinalis* L. Scop, followed by giant foxtail, *Setaria faberi* R. A. W. Herrm, witchgrass, *Panicum capillare* L., woolly cupgrass, *Eriochloa gracilis*, and green foxtail, *Setaria viridis* L. P. Beauv. Infestation in weeks 4, 5, and 6 supported western corn rootworm survivorship, growth, and development better than infestation at later times. Alternate host phenology may be an important larval movement factor between grassy weeds and transgenic rootworm-resistant maize.

**KEY WORDS** *Diabrotica virgifera*, phenology, alternate host, resistance management

THE WESTERN CORN ROOTWORM, *Diabrotica virgifera virgifera* LeConte, is one of the most economically important insect pests of maize, *Zea mays* L. (Levine and Oloumi-Sadeghi 1991), with an extensive range and abundance in North America (Krysan and Branson 1983). Applications of soil insecticides and aerial application of contact insecticides in the past have led to the development of resistance to cyclodiene, organophosphate, and carbamate insecticides in the western corn rootworm (Ball and Weekman 1963, Meinke et al. 1998, Wright et al. 2000). In addition, years of annual maize and soybean rotation in some parts of the U.S. Corn Belt have resulted in the selection for a behavioral variant of the western corn rootworm that overcomes the maize-soybean rotation management tactic. Damage to rotated (first-year) maize by western corn rootworm has been reported in Illinois (Levine and Gray 1996), Indiana (Sammons et al. 1997), Iowa (Rice and Tollefson 1999), and Michigan (DiFonzo

1998). These populations lay eggs in maize fields as well as outside the maize fields (Levine et al. 2002).

Because western corn rootworm has exhibited a plasticity to circumvent most tactics used for managing this pest, new management tactics must be developed and implemented. The latest tool is the deployment of transgenic maize expressing the Cry3Bb1 endotoxins from *Bacillus thuringiensis* Berliner (*Bt*) (Vaughn et al. 2005). The demonstrated ability of corn rootworms to develop resistance to insecticides and overcome the barriers of crop rotation shows the need for a rigorous and cautious insect resistance management (IRM) program. Without such a program, any benefits to farmers and/or the environment from the use of transgenic corn could be fleeting. IRM refers to practices aimed at increasing the time required for insect pests to become resistant to a pesticide.

Several interest groups have expressed concern that widespread planting of transgenic plants could hasten the development of resistance to pesticidal *Bt* endotoxins. Because of this concern, the Environmental Protection Agency (EPA 2003) mandates that all registrants submit an IRM plan before registration of a *Bt* crop. There are a number of research areas for which more information is needed to define an optimal IRM plan for rootworm-resistant transgenic corn,

This article reports the results of research only. Mention of a proprietary product does not constitute endorsement or recommendation of its use by the USDA or the University of Missouri.

<sup>1</sup> Department of Entomology, 1–31 Agriculture Building, University of Missouri, Columbia, MO 65211.

<sup>2</sup> Corresponding author, e-mail: clarkth@missouri.edu.

<sup>3</sup> USDA-ARS, Plant Genetics Research Unit, 205 Curtis Hall, University of Missouri, Columbia, MO 65211.

including the role of alternate hosts in population dynamics and biology of the western corn rootworm.

Branson and Ortman (1967) reported the first successful rearing of the western corn rootworm on the roots of plants other than maize, and recently, Clark and Hibbard (2004), Oyediran et al. (2004), and Wilson and Hibbard (2004) showed larval development to at least second instar on 50 of the 60 species evaluated. Oyediran et al. (2005) showed that significantly more adults emerged from mixtures of Cry3Bb1 transgenic corn with weeds than from weeds or transgenic corn alone. These results indicate that the presence of grassy weeds potentially impacts the production of western corn rootworm adults. However, timing of weed availability and egg hatches could have a large impact. Studies conducted by Apple and Patel (1963) and Strnad and Bergman (1987) suggested that corn rootworm larvae prefer newly developed roots. The objective of our study was to document the impact of host plant phenology on the development and survivorship of western corn rootworm larvae on several host plant species.

### Materials and Methods

**Plant Material and Insects.** The study was conducted in a greenhouse on the University of Missouri-Columbia campus in 2003. The experimental design was a randomized complete block split-split-plot in space and time with five replications. The main plot consisted of six plant species: maize, *Zea mays* L. hybrid P3394 (Pioneer Hi-Bred, Johnston, IA); large crabgrass, *Digitaria sanguinalis* L. Scop; giant foxtail, *Setaria faberi* R.A.W. Herrm; witchgrass, *Panicum capillare* L.; woolly cupgrass, *Eriochloa gracilis* Kunth; and green foxtail, *Setaria viridis* L. P. Beauv. All species except maize were purchased from Valley Seed Service (Fresno, CA). The subplot consisted of eight successive weekly infestations (4–11 wk after planting) and the sub-subplot was comprised of three larval recovery sampling times (7, 14, and 21 d after infestation) to examine larval development. A total of 720 pots were planted: 1 pot for each of the three sample times, for each of the eight infestation times, for each of the six species, and replicated five times.

The plant growth medium was comprised of soil that had been steam-sterilized for 45 min in an autoclave (Amsco Eagle model 2051; Lancer Medical Services, Azusa, CA) and mixed with a peat-based medium (Promix; Premier Horticulture LTEE, Quebec, Canada) in a ratio of 2:1. Plant growth medium was put into 10-liter pots (model confidence limits-600; Hummert International, St. Louis, MO) followed by planting. Each pot had five openings at the bottom that were fitted with a fine (114- $\mu$ m openings) stainless steel mesh (TWP, Berkley, CA) mesh to prevent larval escape (Clark and Hibbard 2004). Planted pots were arranged randomly on greenhouse benches by subplots, each with columns of eight pots that corresponded to infestation time and rows that corre-

sponded to sample date (order was randomized from subplot to subplot).

On germination, seedlings were watered as necessary, fertilized every 2 wk with Peter's fertilizer (20-20-20 NKP; United Industries, St. Louis, MO) at a rate of 0.6 g/liter of water, and grown under 14:10 (L:D) h, with natural light supplemented by 1,000-W sodium bulbs (GE Lighting, Cleveland, OH) and a temperature of  $25 \pm 2^\circ\text{C}$  as monitored with a temperature recorder (model SL 4350 C7C; Dickson Co., Addison, IL).

The larvae used in this study were from the non-diapausing western corn rootworm colony we maintain that was originally obtained from the Northern Grain Insects Research Laboratory, USDA-ARS laboratory, Brookings SD. Larvae from this nondiapausing colony have provided root feeding damage on corn under field conditions that was not significantly different from diapausing western corn rootworm larvae in direct field comparisons (Hibbard et al. 1999). The average head capsule width and dry weight of a subsample of 100 neonate western corn rootworm larvae were determined before infestation and used in subsequent data analysis. From the 4th week through the 11th week after planting, randomly assigned subsamples of each host plant species were infested weekly with 15 neonate western corn rootworm larvae per pot. The infestations were conducted by transferring the neonates to the pots using a moistened fine camel hair brush.

**Data Collection.** After each infestation time, larvae were allowed to feed and then recovered 7, 14, and 21 d after each respective infestation using Tullgren funnels fitted with 60-W light bulbs and collecting jars half-filled with tap water. Each respective larval recovery date was comprised of five pots per alternate host species. Jars were checked after 2 and 4 d in the Tullgren funnels. All recovered larvae were transferred to 95% ethanol, counted, and measured for head capsule width. Each larva from all samples (including the initial neonate subsamples) was measured for head capsule width using an ocular micrometer (10x/21; Wild Co., Heerbrugg, Switzerland) mounted on a microscope (M3Z; Wild Co.). After head capsule measurements were completed, larvae were placed in scintillation vials and dried in a desiccating oven (Thelco model 16; GCA/Precision Scientific Co., Chicago, IL) at  $90^\circ\text{C}$  for 24 h. The dry weight of larvae was measured using an Electronic Balance (SR-182A; A&D Co., Tokyo, Japan), with total weight for all dried larvae from individual samples (including the initial subsample) being recorded. The average dry weight for each individual larva was calculated by simple division.

**Statistical Analysis.** PROC MIXED of the statistical package SAS (SAS Institute 1990) was used for data analysis. A separate analysis was done for percentage larval recovery, larval weight gain, and change in head capsule width. All data were analyzed as a  $6 \times 8 \times 3$  (plant species  $\times$  infestation date  $\times$  sample date) factorial randomized complete block split-split-plot in space as outlined in Steel and Torrie (1980). The linear

Table 1. ANOVA for percentage larval recovery, dry weight gain, and change in head capsule width

Analysis	Effect	df <sub>1</sub>	df <sub>2</sub>	F	P
Larval recovery	Species	5	20	46.09	<0.0001
	Infestation time	7	162	9.03	<0.0001
	Species × infestation time	35	162	1.38	0.0940
	Sampling date	2	372	2.09	0.1254
	Species × sampling date	10	372	4.51	<0.0001
	Infestation time × sampling date	14	372	2.42	0.0029
	Species × infestation time × date	70	372	1.16	0.1999
Dry weight gain	Species	5	20	16.54	<0.0001
	Infestation time	7	162	8.63	<0.0001
	Species × infestation time	35	162	2.76	<0.0001
	Sampling date	2	372	16.85	<0.0001
	Species × sampling date	10	372	14.98	<0.0001
	Infestation time × sampling date	14	372	2.28	0.0054
	Species × infestation time × date	70	372	1.61	0.0030
Change in head capsule width	Species	5	20	60.98	<0.0001
	Infestation time	7	162	23.84	<0.0001
	Species × infestation time	35	162	1.26	0.1668
	Sampling date	2	372	68.24	<0.0001
	Species × sampling date	10	372	13.59	<0.0001
	Infestation time × sampling date	14	372	1.73	0.0472
	Species × infestation time × date	70	372	0.84	0.8191

df<sub>1</sub>, numerator degrees of freedom; df<sub>2</sub>, denominator degrees of freedom.

statistical model contained the main plot of plant species, the subplot effect of infestation date, the sub-subplot effect of sample date, and all possible interactions of plant species × infestation date × sample date. Replications within plant species served as the denominator of *F* for testing the effects of plant species. Replications within infestation dates and sample dates were used as the denominator of *F* for testing treatment and the interaction of infestation dates × sample date. All other effects used the residual error for the denominator. Beyond the standard analysis of variance (ANOVA), we compared the means for the interactions of species and infestation time that were significant ( $\alpha = 0.05$ ). This was done with the LS MEANS output from PROC MIXED using a least significance difference (LSD) test. Although untransformed data are shown in the tables, percentage data were transformed by (arcsine square root of *x*) as suggested by Snedecor and Cochran (1989) for percentage data.

Results

**Larval Survivorship.** Larval survivorship, as indicated by mean percentage larval recovery, was significantly impacted by the host plant species, infestation time, the interaction of host plant species × sample date, and the interaction of infestation time × sample date (Table 1). Larval survivorship was highest in maize followed by large crabgrass (Fig. 1). The number of larvae surviving on all host plants from the first three infestations (fourth, fifth, and sixth weeks) was relatively higher compared with those surviving following subsequent infestations except the last infestation in maize (Fig. 1). Sampling date did not have a significant impact on percentage larval recovery (Table 1).

**Larval Growth and Development.** The average dry weight of neonate western corn rootworm larvae was determined to be 9.298  $\mu\text{g}$  from the pooled subsample of 100 neonate larvae. The average dry weight gain per larva was significantly impacted by host plant species, infestation time, sampling date, and all possible interactions (Table 1). In general, larvae gained more weight during early infestation times (4–6 wk after planting) than they did during late infestations. This difference was significant only within maize and large crabgrass (Table 2). Average weight gain was significantly greater on maize than on all other species and was significantly greater on large crabgrass than on woolly cupgrass or green foxtail (Table 2). Among the grassy weed species, larval weight gain was highest in large crabgrass followed by giant foxtail, witchgrass, woolly cupgrass, and green foxtail, in descending order (Table 2).

The average head capsule width of neonate western corn rootworm larvae was determined to be  $0.220 \pm 0.002$  mm from the subsample of 100 larvae. Increase in mean head capsule width was significantly affected by host plant species, infestation times, sampling dates, host plant species × sampling date, and infestation time × sampling date (Table 1). The interaction between host plant species and infestation time, and the overall interaction between host plant species, infestation time, and sampling date, did not have a significant impact on head capsule width (Table 1). The increase in mean head capsule width generally was highest during the early stages of plant growth (the first few infestations) and gradually declined with infestation time (Fig. 2). Increase in larval head capsule width among species was highest in maize followed by large crabgrass (Fig. 2).

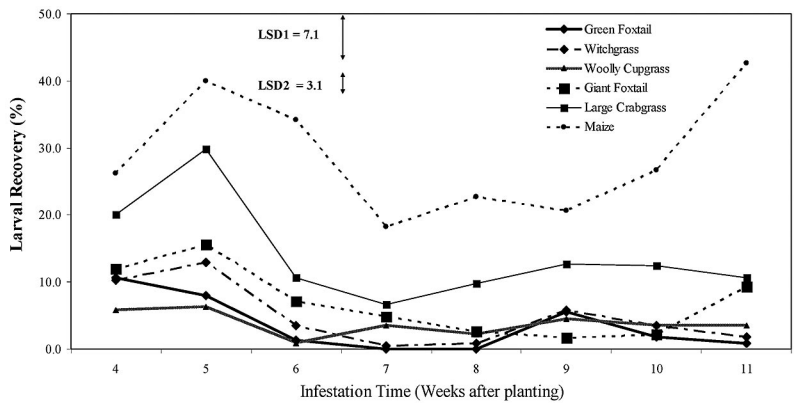


Fig. 1. Percentage larval recovery from six host plant species in different greenhouse pots infested at one of eight weekly infestation times with 15 neonate western corn rootworm larvae. LSD1 is the LSD in percentage larval recovery between host plant species within an infestation date. LSD2 is the LSD in percentage larval recovery between infestation times within a plant species. The three sampling dates for each infestation time were pooled and averaged because there was no significance among them.

Discussion

The commercial release of transgenic maize with resistance to rootworms incorporated with glyphosate tolerance is now a reality, and maize producers in the United States and Canada have a new tool at their disposal to combat the corn rootworm complex. Branson and Ortman (1967, 1970), Clark and Hibbard (2004), and Wilson and Hibbard (2004) showed that many grassy weed species found in or around cornfields are able to support the growth and development of western corn rootworm larvae. Larval development on grassy corn field weeds has potentially important implications regarding the development of appropriate resistance management strategies because larvae could move from weeds to transgenic maize after the weeds are removed or become phenologically less appealing. Results from this study document that host plant phenology could significantly impact survivorship, growth, and development of western corn rootworm larvae and thus support the supposition that host plant phenological changes and subsequent plant nutritional status changes could encourage movement of larvae from weeds to trans-

genic maize. The possibility of such larval movements taking place is supported by findings in a recent study by Hibbard et al. (2004) in which they concluded that western corn rootworm plant-to-plant movement was motivated by a search for food.

This study documents that plant age significantly impacts survivorship, growth, and development of western corn rootworm larvae as indicated by the effect of infestation time on the response variables. Weight gain was reduced and head capsule width increased more slowly when older host plants were infested compared with those that were infested 4–6 wk after planting (Figs. 1 and 2). For example, it is likely that the increase in larval recovery from the last maize infestation time (Fig. 1) was caused by enhanced maize root volume but reduced weight gain (Table 2), and significantly smaller head capsule widths (Fig. 2) compared with larvae recovered from the earlier maize phenologies indicates a loss in nutritional value for developing rootworm larvae as the plants aged. This shows that, although older hosts do support survivorship, they are not as suitable to sustain robust larval growth (weight gain) and de-

Table 2. Average weight gain (dry weight in  $\mu\text{g} \pm \text{SEM}$ ) of larvae recovered from six host plant species in different greenhouse pots infested at one of eight weekly infestation times (I.T.) with 15 neonate western corn rootworm larvae\*

I.T.	Green foxtail	Witchgrass	Cupgrass	Giant foxtail	Large crabgrass	Maize	I.T. main effects
Fourth	49.00 $\pm$ 24.09aC	56.49 $\pm$ 24.54aBC	25.01 $\pm$ 6.89aC	58.63 $\pm$ 20.12aBC	129.64 $\pm$ 49.36aB	479.48 $\pm$ 263.27aA	133.04 $\pm$ 28.07a
Fifth	13.78 $\pm$ 1.00aC	57.55 $\pm$ 24.93aBC	26.37 $\pm$ 13.31aBC	27.68 $\pm$ 10.69aBC	96.07 $\pm$ 39.40aB	372.94 $\pm$ 207.99bA	99.07 $\pm$ 20.43b
Sixth	5.09 $\pm$ 3.25aC	46.68 $\pm$ 36.80aBC	12.09 $\pm$ 9.20aC	197.10 $\pm$ 181.89aB	37.18 $\pm$ 15.04bBC	273.49 $\pm$ 158.17cA	95.27 $\pm$ 34.25b
Seventh	0.00 $\pm$ 0.00aB	46.68 $\pm$ 36.80aBC	13.97 $\pm$ 8.82aB	15.23 $\pm$ 6.53aB	32.32 $\pm$ 16.85bB	145.21 $\pm$ 66.00deA	34.67 $\pm$ 10.23c
Eighth	0.00 $\pm$ 0.00aB	1.38 $\pm$ 1.38aB	7.19 $\pm$ 3.90aB	4.25 $\pm$ 4.25aB	55.32 $\pm$ 32.60bB	168.78 $\pm$ 99.05dA	39.61 $\pm$ 14.00c
Ninth	7.17 $\pm$ 7.67aB	11.81 $\pm$ 11.41aB	14.76 $\pm$ 0.78aB	2.44 $\pm$ 5.37aB	40.89 $\pm$ 16.70bAB	111.87 $\pm$ 49.35deA	34.07 $\pm$ 8.12c
Tenth	6.85 $\pm$ 1.36aB	8.19 $\pm$ 1.57aB	7.52 $\pm$ 4.79aB	5.52 $\pm$ 1.38aB	49.33 $\pm$ 30.86bAB	82.19 $\pm$ 50.13eA	26.60 $\pm$ 6.05c
Eleventh	3.43 $\pm$ 1.80aB	3.21 $\pm$ 1.65aB	8.19 $\pm$ 2.35aB	5.08 $\pm$ 5.08aB	19.67 $\pm$ 7.38bB	103.09 $\pm$ 49.48deA	23.78 $\pm$ 5.65c
SME <sup>a</sup>	10.83 $\pm$ 2.60C	23.78 $\pm$ 6.65BC	14.34 $\pm$ 2.76C	40.89 $\pm$ 23.44BC	58.09 $\pm$ 8.48B	220.77 $\pm$ 27.76A	

\* Data from three sample dates (7, 14, and 21 d) for each infestation time were combined and averaged. Means in the same row followed by the same capital letter and means in the same column followed by the same lowercase letter are not significantly different according to Fisher's protected LSD test ( $\alpha = 0.05$ ).

<sup>a</sup> Species main effects.



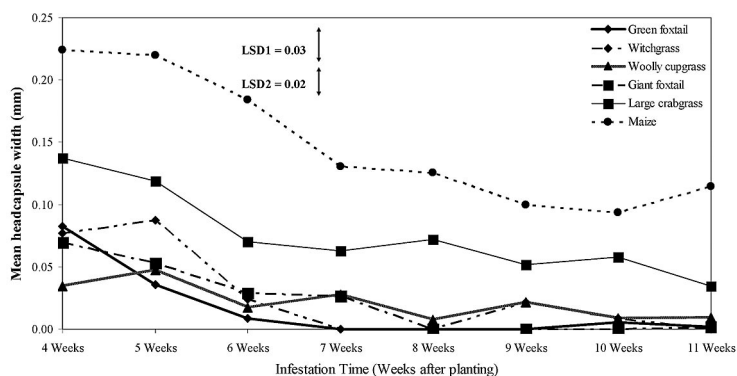


Fig. 2. Mean head capsule width of larvae from six host plant species in different greenhouse pots infested at one of eight weekly infestation times with 15 neonate western corn rootworm larvae. LSD1 is the LSD in head capsule width between host plant species within an infestation date. LSD2 is the LSD in head capsule width between infestation times within a plant species. The three sampling dates for each infestation time were pooled and averaged.

velopment (as determined by increase in head capsule width, which is indicative of development to advanced larval instars). Alternative host plants in the field might support early western corn rootworm larval survival and growth but not necessarily subsequent larval development if only older hosts are available. In related field and greenhouse studies with maize, western corn rootworm larvae could establish on late phenology maize plants, but adult emergence was greatly reduced (B.E.H., unpublished data).

Studies on the feeding behavior of many insects have shown larval feeding changes with seasonal changes in plant nutrients. The nutritional quality of plants to herbivorous insects varies genetically, temporally, and spatially (Whitham 1981, Waltz and Whitham 1997, Lawrence et al. 2003). Seasonal changes in plant nutritional quality are a major source of variation on the temporal scale and have been shown to affect the population dynamics of a broad range of phytophagous insects (Feeny 1970, Kearsley and Whitham 1989). The plant phenological age hypothesis predicts that phytophagous insects should prefer and perform better on phenologically young plants than on old plants because plant nutritional quality decreases with plant phenological age (Scheirs et al. 2002).

As a general rule, the nutrient level, i.e., nitrogen and water, decreases while the non-nutrient content and leaf toughness increase with plant phenological age (Feeny 1970). Three types of processes influence the seasonal changes in plant nutritional quality: ontogenetic processes such as maturation (Kearsley and Whitham 1989), physiological processes such as senescence (Kearsley and Whitham 1989), and a broad range of abiotic (temperature, rainfall) or biotic (herbivores, diseases) environmental variables (Karban 1990).

Feeny (1970) reported that concentration in the spring of feeding by winter moth caterpillars, *Operophtera brumata* L., and other species of Lepidoptera on oak trees in England is believed to be related to seasonal changes in the texture and chemical composition of the leaves. Early feeding was seen to coincide

with maximum leaf protein content and minimum leaf sugar content, which suggested that availability of nitrogen, rather than of carbohydrate, may be a limiting factor for spring-feeding larvae. Riipi et al. (2002) studied seasonal changes in birch leaf chemistry. They reported that co-occurring changes in physical leaf traits and concentrations of several compounds indicated a seasonal decline in foliage suitability for herbivores. Concentrations of birch leaf proteins and free amino acids declined through the growing season and individual sugars showed variable seasonal patterns. While a detailed study examining the nutritional qualities and composition of roots for western corn rootworm development has not been reported, Stavisky and Davis (1997) provided anecdotal evidence to this effect where they observed that western corn rootworm larvae not only prefer newly developed roots (Strnad and Bergman 1987) but may also require younger roots for establishment. It is possible that a similar scenario occurred in the roots of the grassy weeds evaluated in this study but additional experimentation would be required to confirm this scenario.

Percentage larval recovery, head capsule width increase, and larval weight gain differed within and between the various host plant species. This indicates that plant species also significantly impacts larval survivorship, growth, and development. Certain weed species in cornfields could have more serious implications for potential insect resistance management than others. For example, on average, more larvae were recovered from large crabgrass than other grass species, except maize, and they generally gained more weight and increased in head capsule width more as well. This implies that in a corn field, where suitable grassy weeds sometime abound, establishment and development of significant numbers of corn rootworm larvae may occur on species other than maize, and the larvae could move on to transgenic maize, once circumstances in their microenvironment change through weed removal or phenological changes. Because larger larvae are not controlled by the Cry3Bb1 toxin as expressed in current hybrids (EPA 2002), it is

suspected that a significant number of such larvae could withstand the *Bt* toxin in the transgenic maize. The long-term implications of such movement for transgenic events that are not high dose, such as Cry3Bb1, have yet to be determined.

### Acknowledgments

We thank I. Oyediran, T. Wooldridge, K. Moore, A. Antonio, and M. Higdon for assistance at various stages of this project, M. Gacii and B. Gacii for helping with the maintenance of the plants and with data entry, and M. Ellersieck for assistance with statistical analysis and constructive comments on earlier versions of this manuscript. The project was funded, in part, by the USDA CSREES NRI CGP Project Award 2002-35316-12282 and the University of Missouri, Agricultural Experiment Station.

### References Cited

- Apple, J. W., and K. K. Patel. 1963. Sequence of attack by northern corn rootworms on the crown roots of corn. *Proc. N. Cent. Br. Entomol. Soc. Annu. Mtg.* 18: 80–81.
- Ball, H. J., and G. T. Weekman. 1963. Differential resistance of corn rootworms to insecticides in Nebraska and adjoining states. *J. Econ. Entomol.* 56: 553–555.
- Branson, T. F., and E. E. Ortman. 1967. Host range of larvae of the western corn rootworm. *J. Econ. Entomol.* 60: 201–203.
- Branson, T. F., and E. E. Ortman. 1970. The host range of larvae of the western corn rootworm: further studies. *J. Econ. Entomol.* 63: 800–803.
- Clark, T. L., and B. E. Hibbard. 2004. A comparison of non-maize hosts to support western corn rootworm (Coleoptera: Chrysomelidae) larval biology. *Environ. Entomol.* 33: 681–689.
- DiFonzo, C. D. 1998. Special corn rootworm issue. *Mich. State Univ. Ext. Crop Advisory Team Alert.* 13: 12.
- Environmental Protection Agency [EPA]. 2002. Environmental Protection Agency Scientific Advisory Panel: corn rootworm plant-incorporated protectant non-target insect and insect resistance management issues, Part B: insect resistance management issues. <http://www.epa.gov/scipoly/sap/2002/august/august2002final.pdf>.
- Environmental Protection Agency [EPA]. 2003. New corn pest control approved by EPA can lead to reduced pesticide use. <http://yosemite.epa.gov/opa/admpress.nsf/>.
- Feeny, P. 1970. Seasonal changes in oak leaf tannins and nutrients as a cause of spring feeding by winter moth caterpillars. *Ecology.* 51: 565–581.
- Hibbard, B. E., B. D. Barry, L. L. Darrah, J. J. Jackson, L. D. Chandler, L. K. French, and J. A. Mihm. 1999. Controlled field infestations with western corn rootworm (Coleoptera: Chrysomelidae) eggs in Missouri: Effect of egg strains, infestation dates, and infestation levels on corn root damage. *J. Kans. Entomol. Soc.* 72: 214–221.
- Hibbard, B. E., M. L. Higdon, D. P. Duran, Y. M. Schweikert, and M. R. Ellersieck. 2004. Role of egg density on establishment and plant-to-plant movement by western corn rootworm larvae (Coleoptera: Chrysomelidae). *J. Econ. Entomol.* 97: 871–882.
- Karban, R. 1990. Herbivore outbreaks on only young trees: testing hypotheses about aging induced resistance. *Oikos.* 59: 27–32.
- Kearsley, M. J. C., and T. G. Whitham. 1989. Developmental changes in resistance in herbivory: implications for individuals and populations. *Ecology.* 70: 422–434.
- Krysan, J. L., and T. F. Branson. 1983. Biology, ecology and distribution of *Diabrotica*, pp. 144–150. *In* D. T. Gordon, J. K. Knoke, L. R. Nault, R. M. Ritter (eds.), *Proceedings of Maize Virus Disease Colloquium and Workshop*. Ohio Agricultural Research and Development Center, Wooster, OH.
- Lawrence, R., B. M. Potts, and T. G. Whitham. 2003. Relative importance of plant ontogeny, host genetic variation, and leaf age for a common herbivore. *Ecology.* 84: 1171–1178.
- Levine, E., and H. Oloumi-Sadeghi. 1991. Management of diabroticite rootworms in corn. *Annu. Rev. Entomol.* 36: 229–255.
- Levine, E., and M. E. Gray. 1996. First-year corn rootworm injury: east central Illinois research progress to time and recommendations for 1996, pp. 3–13. *In* *Proceedings of the 1996 Illinois Agricultural Pesticide Conference*. Cooperative Extension Service, University of Illinois at Urbana-Champaign, Champaign, IL.
- Levine, E., J. L. Spencer, S. A. Isard, D. W. Onstad, and M. E. Gray. 2002. Adaptation of the western corn rootworm to crop rotation: evolution of a new strain in response to a management practice. *Am. Entomol.* 48: 94–107.
- Meinke, L. J., B. D. Siegfried, R. J. Wright, and L. D. Chandler. 1998. Adult susceptibility of Nebraska western corn rootworm (Coleoptera: Chrysomelidae) populations to selected insecticides. *J. Econ. Entomol.* 91: 594–600.
- Oyediran, I. O., B. E. Hibbard, and T. L. Clark. 2004. Prairie grasses as hosts of the western corn rootworm (Coleoptera: Chrysomelidae). *Environ. Entomol.* 33: 1497–1504.
- Oyediran, I. O., B. E. Hibbard, and T. L. Clark. 2005. Beetle emergence from weedy Cry3Bb1 rootworm-resistant transgenic corn. *J. Econ. Entomol.* 98: 1679–1684.
- Rice, M. E., and J. Tollefson. 1999. Corn rootworms and lodged first-year corn. *Iowa State University Extension, Integrated Crop Management*, IC-482(22).
- Riipi, M., V. Ossipov, K. Lempa, E. Haukioja, J. Koricheva, S. Ossipova, and K. Pihlaja. 2002. Seasonal changes in birch leaf chemistry: are there trade-offs between leaf growth and accumulation of phenolics? *Oecologia (Berl.)*. 130: 380–390.
- Sammons, A. E., C. R. Edwards, L. W. Bledsoe, P. J. Boeve, and J. J. Stuart. 1997. Behavioral and feeding assays reveal a western corn rootworm (Coleoptera: Chrysomelidae) variant that is attracted to soybean. *Environ. Entomol.* 26: 1336–1342.
- SAS Institute. 1990. SAS/STAT user's guide, version 6, 4th ed., vol. 2. SAS Institute, Cary, NC.
- Scheirs, J., L. Debruyne, and R. Verhagen. 2002. Seasonal changes in leaf nutritional quality influence grass miner performance. *Ecol. Entomol.* 27: 84–93.
- Snedecor, G. W., and W. G. Cochran. 1989. *Statistical methods*, 8th ed. Iowa State University Press, Ames, IA.
- Stavisky, J., and P. M. Davis. 1997. The effects of corn maturity class on western corn rootworm (Coleoptera: Chrysomelidae) phenology. *J. Kans. Entomol. Soc.* 70: 261–271.
- Steel R. G. D., and J. H. Torrie. 1980. *Principles and procedures of statistics: a biometric approach*, 2nd ed. McGraw-Hill, New York.
- Strnad, S. P., and M. K. Bergman. 1987. Distribution and orientation of western corn rootworm (Coleoptera: Chrysomelidae) larvae in corn roots. *Environ. Entomol.* 16: 1193–1198.

- Vaughn, T. T., T. Cavato, G. Brar, T. Coombe, T. DeGooyer, S. Ford, M. Groth, A. Howe, S. Johnson, K. Kolacz, C. Pilcher, J. Purcell, C. Romano, L. English, and J. Pershing. 2005. A method of controlling corn rootworm feeding using a *Bacillus thuringiensis* protein expressed in transgenic maize. *Crop Sci.* 45: 931–938.
- Waltz, A. M., and T. G. Whitham. 1997. Plant development affects arthropod communities: opposing impacts of species removal. *Ecology*. 78: 2133–2144.
- Whitham, T. G. 1981. Individual trees as heterogeneous environments: adaptation to herbivory or epigenetic noise, pp. 9–27. In R. F. Denno and H. Dingle (eds.), *Insect life history patterns*. Springer, New York.
- Wilson, T. A., and B. E. Hibbard. 2004. Host suitability of nonmaize agroecosystem grasses for the western corn rootworm (Coleoptera: Chrysomelidae). *Environ. Entomol.* 33: 1102–1108.
- Wright, R. J., M. E. Scharf, L. J. Meinke, X. Zhou, B. D. Siegfried, and L. D. Chandler. 2000. Larval susceptibility of an insecticide-resistant western corn rootworm (Coleoptera: Chrysomelidae) population to soil insecticides: laboratory bioassays of detoxification enzymes, and field performance. *J. Econ. Entomol.* 93: 7–13.

*Received for publication 15 March 2005; accepted 29 August 2005.*

---